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**Environmental Genomic Characterization of the Deep Subsurface
Microorganism *Desulforudis audaxviator***

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PRESENTATION INFORMATION

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SUMMARY

*"In Sneffels Joculis craterem quem delibat Umbra Scartaris Julii intra calendas
descende, **Audax viator**, et terrestre centrum attinges."*

*("Descend, **bold traveller**, into the crater of the jokul of Sneffels, which the shadow of
Scartaris touches before the kalends of July, and you will attain the center of the earth.")*

-- Hidden message deciphered from an Icelandic saga that prompts Professor Lidenbrock to undertake his travels in Jules Verne's "Journey to the Center of the Earth"

A more complete picture of life on Earth, and even life *in* the Earth, has recently become possible through the application of environmental genomics, in which DNA is extracted from an environmental sample and then sequenced and analyzed. The field of environmental genomics is expanding our view of the extent of life on this planet and allowing us to begin to characterize the abilities of microbes that inhabit niches previously thought to be inhospitable, even when isolation of the organisms present in those environments is not possible. A team of researchers is using this approach to study life deep within the crust of the earth, and report here the genome sequence of the dominant organism found in the deep subsurface of South Africa. This organism, which they have named *Desulforudis audaxviator* in honor of its bold travels, its rod-like morphology, and its apparent use of sulfate instead of oxygen for the generation of energy, demonstrates a successful strategy for life in the deep subsurface, and may prove to be similar to organisms in equivalent environments in other regions of the Earth's crust.

The study reported here at the ASM General Meeting in Orlando, Florida, on May 22nd, 2006 has required the abilities of a team of investigators. Tullis C. Onstott from Princeton University led the team that collected the sample, Fred J. Brockman and David E. Culley from the Pacific Northwest National Laboratory isolated the DNA, Alla Lapidus from the DOE Joint Genome Institute led the sequencing efforts, and Eric J. Alm of the Massachusetts Institute of Technology and Eoin L. Brodie, Adam P. Arkin, Terry C. Hazen, and Dylan Chivian from the E.O. Lawrence Berkeley National Laboratory performed the analysis of the genome. This project has received funding from the DOE Genomics:GTL program, as well as from the NASA Astrobiology Institute, as such deep subsurface environments on Earth allow us to understand by analogy environments that may be conducive to life on Mars.

The Gold and Platinum mines of South Africa have provided access to microorganism bearing fluid that emanates from fractures at depths ranging from 0.7 to 3.2 km below the

surface and possibly up to 5 kms. Of the ~280 bacterial and 44 archaeal types identified in the South African mines only 11 bacterial and one archaeal species have been isolated but none of the novel lineages have been successfully enriched. One novel type belonging to the Gram-positive bacteria (a member of the *Firmicutes*) has been found in almost all fracture fluids emanating from depths greater than 1.5 km across the entire Witwatersrand Basin (150 x 300 km), and has never been cultivated, making it a perfect candidate for environmental genomics. In order to better understand this prevalent organism, a genomic analysis was performed on DNA extracted from a borehole water sample collected at 2.8 km depth where analyses indicate that the sparse community is dominated at greater than 93% by the *Desulforudis* type.

The genome content of *Desulforudis audaxviator* supports the geochemical prediction that sulfate respiration (instead of oxygen) is the optimal metabolic strategy in this environment. Genes that are involved in formate oxidation and in the assimilation of carbon monoxide, which are present in abundance, were also found. Additionally, nitrogen is required by all organisms, yet N₂ fixation directly from the atmosphere is an energetically costly process performed by only a fraction of organisms (including some related *Clostridium* species). Consistent with their dominant presence in these environments, a N₂ fixation pathway was identified within the genome. *D. audaxviator* has genes that appear to be associated with organisms that live in hot environments (the temperature where the sample was taken was roughly 60° Celsius), but it is not yet clear whether this organism can tolerate extremely high temperatures. The presence of genes for the formation of the flagellar assembly (swimming apparatus), and the sensory receptors that guide it, implies that this organism is capable of swimming towards conditions that are more hospitable for growth, such as concentrations of nutrients. Undoubtedly, more discoveries about the nature of deep life will soon follow, and it is somewhat fitting that deciphering the genome of *Desulforudis audaxviator* has been an undertaking reminiscent of the efforts of the fictitious Professor Lidenbrock to find his own hidden message to reveal the secrets at the center of the Earth.

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